CLAIMS

That which is claimed is:

1. A method of identifying a nucleic acid encoding a signal sequence, the method comprising:

directionally introducing a cDNA into a vector comprising a nucleic acid encoding a leaderless secretable selection protein to produce a fusion nucleic acid insert in said vector, the fusion nucleic acid encoding a fusion protein;

introducing the vector comprising the fusion nucleic acid into a bacterial cell, said introducing allowing for expression of the fusion protein;

exposing the bacterial cell to a selection medium, wherein said selection medium supports growth of bacteria that secrete the fusion protein; and

determining growth of the bacterial cells in said selection medium;

wherein growth of the bacterial cells in said selection medium indicates that the nucleic acid encodes a signal sequence.

2. The method of claim 1, wherein the vector is a dual expression vector.

3. The method of claim 2, wherein the vector comprises a mammalian promoter and a bacterial promoter.

4. A method of identifying a nucleic acid encoding a signal sequence, comprising:

directionally introducing a cDNA into a vector comprising a nucleic acid encoding a leaderless β-lactamase to produce a fusion nucleic acid insert in said vector, the fusion nucleic acid encoding a fusion protein;

introducing the vector comprising the fusion nucleic acid in a bacterial cell, said introducing allowing for expression of the fusion protein;

exposing the bacterial cell to a selection medium; and

determining growth of the bacterial cells in said selection medium, wherein the selection medium supports growth of bacteria that secrete the fusion protein;

wherein growth of the bacterial cells in said selection medium indicates that the nucleic acid encodes a signal sequence.

5. The method of claim 4, wherein the selection medium is a medium comprising β-lactam antibiotic.

6. The method of claim 5, wherein the selection medium comprises ampicillin.

7. The method of claim 4, wherein the vector is a dual expression vector.

8. The method of claim 7, wherein the vector comprises a mammalian promoter and a bacterial promoter.

9. A method of identifying a cDNA encoding a signal sequence, comprising:

directionally introducing a cDNA into a vector, said vector comprising:

a prokaryotic promoter, a eukaryotic promoter, a multiple cloning site, and a nucleic acid encoding a leaderless secretable selection protein, wherein said introducing results in the formation of a fusion nucleic acid;

introducing the vector comprising the fusion nucleic acid into a bacterial cell;

exposing the bacterial cell containing the cDNA to a selection medium;

determining growth of the bacterial cell in said selection medium, wherein growth of the bacterial cells in said selection medium is indicative of a signal sequence in said cDNA;

introducing the vector identified as comprising a signal sequence into eukaryotic cells;

culturing the transfected eukaryotic cells; and

detecting secretion of the cDNA-selection protein fusion in the cell culture;

wherein the vector expresses a fusion protein encoded by the cDNA and the nucleic acid encoding the selection protein.

10. A method of identifying a cDNA encoding a protein having a signal sequence, comprising:

directionally introducing a cDNA into a vector, said vector comprising a prokaryotic promoter, a eukaryotic promoter, a multiple cloning site, and a nucleic acid encoding a leaderless β-lactamase protein, wherein said introducing results in the formation of a cDNA-β-lactamase fusion nucleic acid;

introducing the vector comprising the fusion nucleic acid into a bacterial cell;

exposing the bacterial cell to a selection medium;

determining growth of the bacterial cell in said selection medium, wherein growth of the bacterial cells in said selection medium is indicative of a signal sequence in said cDNA;

introducing the vector identified as comprising a signal sequence into eukaryotic cells;

culturing the transfected eukaryotic cells; and

detecting secretion of the cDNA-selection protein fusion in the cell culture; wherein the vector expresses a fusion protein encoded by the cDNA and the nucleic acid encoding the selection protein.

11. The method of claim 10, wherein the selection medium is a medium comprising β-lactam antibiotic.

12. The method of claim 11, wherein the selection medium comprises ampicillin.

13. The method of clam 10, wherein the β-lactamase is detected in cell culture using a nitrocefin hydrolysis assay.

14. The method of claim 6, wherein the vector comprises a mammalian promoter and a bacterial promoter.

15. A method of producing a cDNA library enriched for proteins comprising signal sequences, said method comprising:

directionally introducing each of a plurality of cDNAs into a vector, said vector comprising a nucleic acid encoding a leaderless secretable selection protein;

introducing each vector into a bacterial cell to create a library comprising the plurality of cDNAs;

expressing the cDNAs in the bacterial cells; and

selecting bacterial cells containing a cDNA encoding a secreted protein by growth in a selection medium;

wherein the selected bacterial cells are enriched for proteins comprising signal sequences.

16. The method of claim 15, wherein the cDNAs are 5' biased.

17. The method of claim 15, wherein the bacterial cells are subjected to a second round of selection in a selection medium.

18. A high throughput method of identifying a cDNA which encodes a secreted protein, said method comprising:

directionally introducing each of a plurality of cDNAs individually into a vector comprising a nucleic acid encoding a leaderless secretable selection protein, wherein said introducing results in the formation of a cDNA-β-lactamase fusion nucleic acids in a plurality of vectors;

introducing the plurality of vectors into bacterial cells to create a bacterial cell library; and

selecting bacterial cells containing a cDNA encoding a signal sequence by growth in a selection medium;

wherein growth of the bacterial cells in said medium indicates that the cDNA comprises a signal sequence.

19. The method of claim 18, further comprising the steps of isolating the vector from the selected bacterial cells and identifying the sequence of the cDNA.

20. The method of claim 18, wherein the method further comprises determining the sequence of the cDNA inserts.

21. A method for detecting secretion of a protein comprising a signal sequence, said method comprising the steps:

directionally introducing a cDNA encoding a protein into a vector, said vector comprising a nucleic acid encoding a leaderless secretable selection protein, wherein introducing the cDNA into the cell produces a cDNA-selection protein fusion vector;

introducing the protein fusion vector into a bacterial cell;

exposing the bacterial cells containing the nucleic acid fusion to a selection medium; and

determining growth of the bacterial cells in said selection medium;

wherein growth of the bacterial cells indicate that the cDNA encodes a protein comprising a signal sequence.

22. A method for detecting secretion of a protein comprising a signal sequence, said method comprising the steps:

directionally introducing a cDNA encoding a protein into a vector, said vector comprising a nucleic acid encoding a leaderless β-lactamase protein, wherein introducing the cDNA into the cell produces a cDNA- β-lactamase fusion vector;

introducing the fusion vector into a bacterial cell;

exposing the bacterial cells containing the nucleic acid fusion to a selection medium; and

determining growth of the bacterial cells in said selection medium;

wherein growth of the bacterial cells indicate that the cDNA encodes a protein comprising a signal sequence.

23. A vector for identifying a cDNA insert encoding a protein comprising a signal sequence, said vector comprising a prokaryotic promoter, a eukaryotic promoter, a multiple cloning site, and a nucleic acid encoding a leaderless secretable selection protein.

24. The vector of claim 23, wherein the prokaryotic promoter is a bacterial promoter, and wherein the eukaryotic promoter is a mammalian promoter.

25. The vector of claim 23, wherein the secretable selection protein is β-lactamase.

26. The vector of claim 23, wherein the vector is pBK-CMV-leaderless-β-lactamase.